



Serological Evidence of Caprine Arthritis Encephalitis in North Shewa Zone, Ethiopia: Clinical Case Analysis

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Introduction: Caprine arthritis encephalitis (CAE) is a chronic debilitating and economically important viral disease of goats. It is mainly manifested as encephalitis in kids and polyarthritis in adult goats. The present study was conducted to determine the rate of morbidity and mortality due to clinical diseases attributed to infection by *Caprine arthritis encephalitis virus* (CAEV) and to determine the serological status of CAEV in goat in North Shewa, Ethiopia.

Methods: A cross-sectional serological study and a longitudinal clinical case study were conducted. A total of 257 serum samples have been collected from apparently health and clinical cases attributed to CAE infection and tested with the usage of indirect enzyme-linked immunosorbent assays to screen antibodies against CAE. Records have been statistically analyzed by using the chi-square test.

Results: During five consecutive years of longitudinal clinical study, a total of 195 clinical diseases of chronic pneumonia, nerve problems, clinical mastitis, and arthritis occurred with prevalence of 99 (50.8%), 57 (29.2%), 27 (13.9%), and 12 (6.2%), respectively. Chronic pneumonia was the highest cause of goat morbidity (50.8%) and mortality (100.0%). Of the total samples tested from clinical cases, 7 (58.3%) were sero-positive for the presence of antibodies against CAEV. The overall seroprevalence of CAE was 4.7%. There has been a significant difference ($p < 0.05$) in sero-positivity among management system, breeds, and age groups of goats. However, there was no significant variation in sero-positivity between the sexes ($p > 0.05$) of goats.

Conclusion: This finding indicates that CAEV infection exists in the goat flocks in examined localities in Ethiopia. This disease poses serious animal health problems that constrain production with the presence of apparent clinical signs. Further investigations need to be done to explore the seroconversion of CAEV in small ruminants and the associated factors to plan an appropriate eradication program and prevent transmission.

Keywords: caprine arthritis encephalitis, goats, seroprevalence

Introduction

Caprine arthritis encephalitis (CAE) is a critical viral disorder of goats due to *Caprine arthritis encephalitis virus* (CAEV), a Lentivirus of the own family Retroviridae.¹ CAEV produces an insidious, persistent, and slowly progressive systemic inflammatory infection,^{2,3} mainly characterized by polyarthritis, interstitial pneumonia, indurative mastitis, and progressive weight loss in adult goats and encephalitis in kids.⁴ Having a protracted incubation length accompanied by a persistent clinical course,⁵ CAEV infects its host for lifestyles as soon as the infection is mounted, in spite of the presence of humoral and cellular mediated immune reaction.⁶ All infected animals grow to be capability transmitters of virus, although maximum of that are typically asymptomatic sub-scientific vendors.⁷ Transmission frequently takes vicinity through ingestion of virus-infected colostrums or goat milk,⁸ irrespective of the presence of maternal antibody,⁹ and much less usually through other routes consisting of direct contact, bodily secretions, and excretions.¹⁰ However, unlike other Lentiviruses, sexual transmission has not but been nicely described for CAEV.¹¹

The diagnosis of the disease involves a combination of clinical manifestation, postmortem examination, and histopathological findings. However, serology is considered as one of the easiest and most efficient ways to diagnose infections caused by CAEV.¹² In practice, the most common serological test adopted for the diagnosis of CAEV infection is enzyme-linked immunosorbent assay (ELISA) and agar gel immunodiffusion (AGID).¹³ However, ELISA test is the most preferred due to the low sensitivity associated with AGID.¹² Thus far, no treatment has come out but for the relief of CAEV infection;¹⁴ therefore early detection of infection using serological diagnostic techniques¹⁵ remains an essential technique for prevention, management, and eradication of CAEV infection.^{14,16}

CAEV is worldwide in distribution.¹⁷ It has been detected in lots of parts of the world considering the fact that its first documentation in goats was in 1974.^{18,19} In advance surveys in Africa documented the proof for small ruminant lentivirus (SRLV) infections in Algeria,²⁰ Morocco,²¹ and Mozambique.²² Also, CAE has been reported in nearby countries; Sudan,⁶ Somalia,¹³ and Kenya.²³ But, to our information, there was no report on the prevalence of CAE both in the goat and sheep population in Ethiopia.²⁴ However, following importation of Boer goat to home country for genetic upgrading, an occurrence of a new case with undefined etiology characterized through regular clinical signs of CAEV has been located in imported unique Boer goats at Ataye Boer nucleus site, Ethiopia. It has been a critical hassle repeatedly visible where 25.5% of goat mortality has been said.²⁵

Signs and symptoms observed at the Boer nucleus site have been indicators of CAEV infection within the flock. Clinically, the goats confirmed dyspnea with heavy breathing and rearing sound, revolutionary pneumonias, chronic thin vs serous nasal discharge with foam on mouth, serous fluid lacrimation with protrusion of the eyeball and dilated pupil, innovative body weight reduction, arthritis, listlessness, torticollis, twisted neck, circling, and a number of kids born and dead after few weeks or months with anxious sign, mastitis and agalactia, decreased, absent milk production, and rough hair coat.^{26,27} The ailment caused reproductive wastage and neonatal disease of goats main to big monetary loss. Therefore, the present study was designed to investigate if CAEV is the causal agent of the observed clinical signs in the goat population of the study areas through determination of its seroprevalence and to determine the morbidity and mortality rate due to clinical diseases; chronic pneumonia, nerve problems, clinical mastitis, and arthritis attributed to infection by CAEV.

Materials and Methods

Description of Study Areas

The study was carried out in two districts of North Shewa zone of Amhara regional state, Ethiopia. Of these districts, Efratanagidim (Laygnawataye, Jeweha negeso and Ataye Boer nucleus site within Laygnawataye kebele) and Kewet (Jimderena gur, yelen and Shewarobit town) were decided on purposively (Figure 1). During the last few years (2013–2019) a number of Boer cross bucks originated from DBARC; Ataye Boer nucleus site had been dispensed to small holders to improve the genetic capacity of the local indigenous goat (central highland goat) breeds. The on-farm study areas were decided on based on retrospective data showing the history of introduction of Boer cross bucks.

Kewet and Efratanagidim are located 200 and 250 km from Addis Ababa, respectively. Geographically, the areas are placed 10°21'0" N and 39°55'60" E with average elevation stages of 1280–1468 meters above sea level (Figure 1). The climate is characterized by bimodal rainfall consisting of the lengthy wet season (June–September), short rainy season (February–May additionally), and dry season (October–January). In 12 months, the average rainfall is 1085–1199 mm.²⁸ They have an average temperature of 25.4–27.0 °C; June is the hottest month of the 12 months. December has the bottom common temperature of the year.

Study Animals and Their Management

Animals used for this study have been pure Boer, central Highland Goat (local breed), 50% Boer (pure Boer cross central Highland Goat) and 25% Boer (50% Boer cross central Highland Goat). Age categorization into younger and adult become decided as defined by Casburn²⁹ for look at animals. For that reason have a look at animals up to one year had been categorized as younger and the relaxation as adult. According to Morris,³⁰ the production system was classified into two primarily based on management systems of animal owners in the have a look at study areas. Animals owned with the

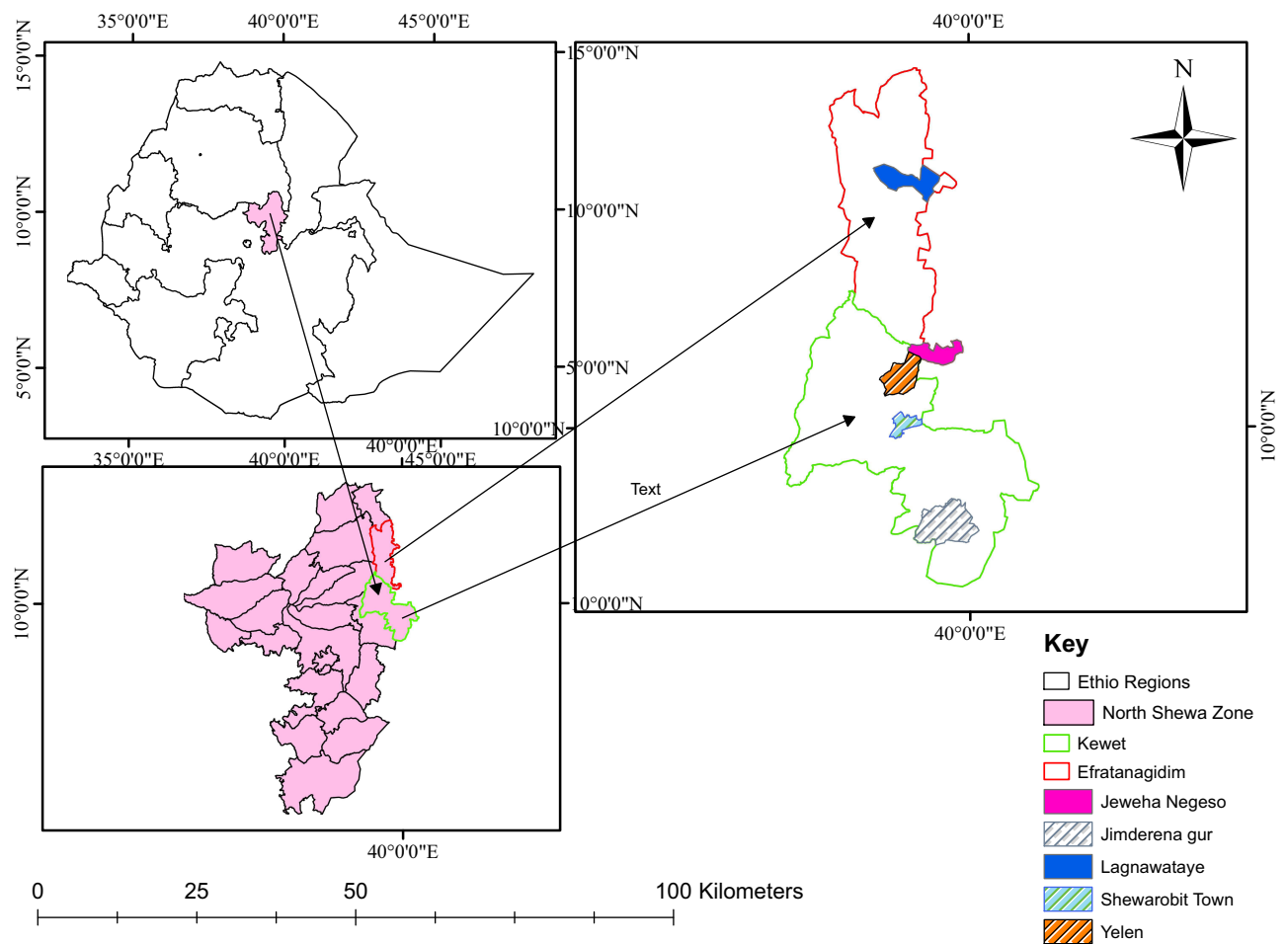


Figure 1 Map of the study areas.

aid of man or woman farmers were managed under a traditional grazing (extensive) system and on station have been under a semi-intensive system. In the extensive system, animals spent all the day browsing and grazing pasture on fallow lands with no extra-supplement and sheltered for the duration of the night time while in semi-extensive they were more supplemented in addition to grazing. The complement consists of advert libitum grass hay, chopped pasture (Napier grass, *Desmodium* species and vetch) and commercial concentrate based on their body weight.³¹

Furthermore, animals controlled under semi-intensive and extensive were treated using anthelmintic drugs (albendazole, tetramisole, ivermectin and triclabendazole) based on the epidemiological cycle of targeted parasite groups and the laboratory findings. They have been also vaccinated against major viral and bacterial infectious diseases which consist of ovine pasteurellosis, sheep and goat pox, peste des petits ruminants' (PPR) and contagious caprine pleura pneumonia (CCPP).³¹

Study Design and Sample Size Determination

Longitudinal Clinical Case Study

According to Humberto et al,⁴ clinical diseases such as chronic pneumonia, arthritis, clinical mastitis and nerve signs attributed to infection by CAEV were considered and analyzed by using five years (September 1, 2014 to August 30, 2019) longitudinal data (both retrospective and prospective) in Ataye Boer nucleus site; Debre-Birhan Agricultural Research Center, Ethiopia.

All goats were clinically examined by assessment of general conditions and gait, as well as inspection and palpation of the extremities joints. Presence of clinical arthritis (swelling and stiffness with lower mobility) was noted bilaterally. Clinical arthritis was diagnosed according to the method described by Bertoni et al.³² Physical examination of clinical mastitis was

carried out by means of traditional methods as described by Birgel.³³ Diagnosis of nerve signs (circling, staggering gait, torticollis, twisting of neck, paresis, and paralysis) and chronic pneumonia (heavy breathing, coughing and rearing sound, chronic pneumonia, and thin nasal discharge with foam on mouth) were conducted based on clinical examination. Generally, there were a regular follow up of sick animals and types of disease were registered daily in case recording book.

Cross-Sectional Serological Study

Cross-sectional seroprevalence study was carried out from August to October, 2019. The study animals have been sampled using simple random sampling techniques in all districts of the study areas. To our information, there was no report on the prevalence of CAE both in the goat and sheep population in Ethiopia²⁴ and this examine become taken into consideration as a first investigation in the country. To decide the seroprevalence of CAE within the study flocks, an appropriate sample size was computed with the usage of the method cautioned by Thrusfield.³⁴ In case there was no previous study and the expected prevalence was 50% and targeted population size was 650.

$$n = \frac{1.96^2 p_{ex}(1-p_{ex})}{d^2} \quad (1)$$

$$n^* = 1/(1/n + 1/N) \quad (2)$$

Where: n = required sample size while n is greater than 10% of the total population size, P_{ex} = expected prevalence, d = desired absolute precision, 1.962 = the value of z at 95% confidence level, n^* = actual required simple size calculated, and N = population size.

Accordingly, the required sample size was 242 goats, but to increase the precision and accuracy, we collected 257 samples.

Sample Collection and Serological Examination

Examined animals above six months of age have been used for the sample collection. Blood samples have been gathered in accordance with standard procedures directly from each selected animal's jugular vein the usage of simple vacutainer tubes and sterile needles and allowed to clot for 1–2 h at room temperature; saved horizontally overnight at 4 °C and finally, the serum become separated from the clot. The separated serum was labelled and become stored in a refrigerator (–20 °C) until tested.

The serum was analyzed at National Animal Health Diagnostic and Investigations Centre, Sebeta, Ethiopia. All sera were tested with the *CAEV/MVV* Total Ab (IDEXX Switzerland AG, Liebefeld-Bern, Switzerland) for the presence of antibody against CAEV. The sensitivity and specificity of the ELISA test kit as provided by the manufacturer was 98.6% and 99.3%, respectively. Sera were tested and scored according to the manufacturer's instruction.

Sample to positive (SP) % values were calculated and interpreted as follows: sera with SP% values below 30 were considered negative, sera with SP% values between 30 and 40 doubtful, and sera with SP% values above 40 were classified as positive. The mean SP% values are reported in the text with the corresponding standard deviations (SD).

Data Management and Analysis

Data collected during sampling and laboratory result had been entered in a MS-Excel spreadsheet and analyzed by means of the use of SPSS software program (version 20). The seroprevalence became determined by way of dividing the overall quantity of positive samples for the presence of antibody against CAEV by the overall variety of samples analyzed.³⁵ Proportions were calculated for seroprevalence vis-à-vis fixed factors that included only goats' sex, age, breed, management system, and village. Difference in seroprevalence amongst intercourse, age, management system, and breed of goats became analyzed with the usage of chi-square evaluation and in comparison consistent with the fixed factors and confidence limit of less than 5% was used to indicate a significant level ($p < 0.05$).

Results

Longitudinal Clinical Case Study

Clinical case analysis attributed to infection by CAEV with fixed factors was conducted. Of total 195 clinical diseases, chronic pneumonia and arthritis were the highest and the lowest occurrences with relative prevalence of 50.8% and 6.2%, respectively. Relatively, adult (72.3%) and female (65.1%) were more prone to infection than respective young (35.9%) and male (34.9%), respectively. Central highland goats (17.4%) were relatively tolerant to infection than other breeds (Table 1).

Chronic pneumonia was relatively the highest cause of goat morbidity and mortality with overall relative rates of 50.8% and 100.0%, respectively. All animal manifest chronic pneumonia and nerve problems were died and clinical mastitis and arthritis cases were out of breeding due to poor production (low milk production and progressive weight loss) (Table 2).

From the total of 116 serum sample collected from Ataye Boer nucleus site, 12 were from clinical cases (chronic pneumonia, clinical mastitis, and arthritis attributed to infection by CAEV). But, there was no chance of collecting samples from cases of nerve problems at Ataye boer nucleus site and also no case samples from all other study areas. Of the total samples tested from clinical cases, 7 (58.3%) were sero-positive for the presence of antibodies against CAEV. From the total of 12 clinical diseases of chronic pneumonia, clinical mastitis, and arthritis, sero-positivity occurred with relative prevalence of 80, 33.3, and 50%, respectively (Table 3). Such sero-positivity of clinical case to CAE antibody confirms the presence of the disease in the study flock and in the country.

Cross-Sectional Serological Results

From the total samples tested, 12 (4.7%) were positive for antibodies against CAEV. In this study the highest sero-positivity rate was recorded in goats at Ataye Boer nucleus site with prevalence of 10 (8.6%) but no antibodies were

Table 1 Statistical Analysis of Different Clinical Diseases Attributed to Infection by CAEV with Various Fixed Risk Factors at Ataye Boer Nucleus Site

Variables		Clinical Diseases				Total (%)	χ^2 -value	p-value
		Chronic Pneumonia (%)	Nerve Problem (%)	Clinical Mastitis (%)	Arthritis (%)			
Age	Young	25 (12.8)	24 (12.3)	–	5 (2.6)	54 (27.7)	17.718	0.001
	Adult	74 (37.9)	33 (16.9)	27 (13.8)	7 (3.6)	141 (72.3)		
Breed	Boer	38 (19.5)	14 (7.2)	6 (3.1)	4 (2.1)	62 (31.8)	25.385	0.000
	Boer × CHG*	54 (27.7)	27 (13.8)	10 (5.1)	8 (4.1)	99 (50.8)		
	CHG*	7 (3.6)	16 (8.2)	11 (5.6)	0 (0.0)	34 (17.4)		
Sex	Female	69 (35.4)	27 (13.8)	27 (13.8)	4 (2.1)	127 (65.1)	28.624	0.000
	Male	30 (15.4)	30 (15.4)	–	8 (4.1)	68 (34.9)		
Grand total		99 (50.8)	57 (29.2)	27 (13.9)	12 (6.2)	195 (100.0)		

Abbreviation: *CHG, central highland goat.

Table 2 Rate of Goat Morbidity and Mortality Due to Different Clinical Signs of CAE in Ataye Boer Nucleus Site

Clinical Diseases	No. of Morbidity (%)	No. of Mortality (%)
Chronic pneumonia	99 (50.8)	99 (100.0)
Nerve problems	57 (29.2)	57 (100.0)
Clinical mastitis	27 (13.9)	0 (0.0)
Arthritis	12 (6.2)	0 (0.0)
Total	195 (100.0)	156 (80.0%)

Table 3 Apparent Seroprevalence of CAE in Active Cases at Ataye Boer Nucleus Site

Clinical Diseases	No. of Serum Sample Collected	No. of Sero-Reactors (%)
Chronic pneumonia	5	4 (80)
Clinical mastitis	3	1 (33.3)
Arthritis	4	2 (50)
Total	12	7 (58.3)

detected in three villages namely Jeweha negeso, Shewarobit town and Yelen. The distribution of sero-positivity to CAEV among the six sites/villages is shown in Table 4.

There was a statistically significant difference in the sero-positivity recorded between age groups ($p < 0.05$); adults were more prone to CAEV infection as compared to young. There was also a statistically significant difference in the sero-positivity among different breeds of goats ($p < 0.05$) in which Pure Boer and 50% Boer were more affected. Also, there was a statistical significant difference in sero-positivity between management/husbandry systems; semi-intensively managed animals were more prone to infection than extensively managed. But, there was no significant difference in sero-positivity between sexes of goats as stated in Table 5.

Table 4 Seroprevalence of CAEV in Goat by I-ELISA in North Shewa, Ethiopia

District	Study Sites	No. of Animals Sampled	No. Positive (%)
Efratanagidim	Ataye Boer nucleus site	116	10 (8.6)
	Laygnawataye	11	1 (9.1)
	Jeweha negeso	45	0 (0.0)
Kewet	Yelen	15	0 (0.0)
	Shewarobit town	17	0 (0.0)
	Jimderena gur	53	1 (1.9)
Total		257	12 (4.7)

Table 5 Sero-Positivity to CAEV Infection in Goat Based on Various Fixed Variables

Variables		No. Sampled	No. Positive (%)	χ^2 -value	p-value
Sex	Female	175	7 (4.0)	0.552	0.458
	Male	82	5 (6.1)		
Age	Adult	155	11 (7.1)	5.170	0.023
	Young	102	1 (1.0)		
Breed	Pure Boer	21	2 (9.5)	8.082	0.044
	50% Boer	75	7 (9.3)		
	25% Boer	43	0 (0.0)		
	CHG*	118	3 (2.5)		
Management system	Semi-intensive	116	10 (8.6)	7.417	0.006
	Extensive	141	2 (1.4)		
Overall		257	12 (4.7)		

Abbreviation: *CHG, central highland goat breed.

Discussions

CAE is a chronic debilitating and economically important viral disease of goats. The disease is mainly manifested as subclinical; however, a small number of the animals develop progressive, untreatable disease that includes encephalitis in kids and polyarthritis in adult goats.³⁶ In addition, the disease decreases lifetime productivity in goats.³⁷

Clinical diseases of chronic pneumonia, nerve problems, clinical mastitis, and arthritis attributed to infection by CAEV were major goat problems, causing high productive and reproductive loss through morbidity and mortality in Ataye Boer nucleus site, which was agreed with different reports.^{4,38–40} In this study, all goats' manifest chronic pneumonia and nerve problems were died; they were not responded to antibiotic therapy. There was no treatment to come out for the relief of clinical cases, since all clinical cases were never responded to antibiotic treatment.¹⁴

Furthermore, clinical mastitis was the principal cause of doe culling due to poor production of milk, which was coinciding with the findings of Gregory et al,³⁸ Lerondelle et al,⁴¹ Lerondelle et al,⁴² and Lara et al,⁴³ and they reported diffuse hardening of the parenchyma of the glands, with some cases of hypotrophy or atrophy of the infected halves. Substitution of gland parenchyma by connective tissue and fibrosis observed in the histopathological examination explains the decrease in milk yield described by Bohland and D'Angelino⁴⁴ in 2005, and the quality of milk produced by animals infected by the virus.⁴⁵ Also, mastitis cases were a serious problem in the study site, causing a high rate of kid mortality due to agalactia.³¹

The serological result of the present study revealed evidence of CAEV infection within goat's population in two districts; Efratanagidim and Kewet of North Shewa, Ethiopia with an overall seroprevalence of 4.7%. The seroprevalence recorded was quite interesting since, to our knowledge, there was no report on the prevalence of CAEV both in the goat and sheep population in Ethiopia.²⁴ The observed sero-positivity of the disease in those study sites that are geographically far from the relatively affected site (on-station) suggest that the disease might have been spread along with the distribution of Boer cross bucks. The lack of published studies on the CAEV situation in Ethiopia makes the present study interesting to understand the epidemiology of CAEV infection in goats in different livestock production contexts in Ethiopia.

The 4.7% prevalence of CAEV in this study was comparable with the reports in India 3.33%,¹² Northern Somalia 6%,¹³ Malaysia 8.8%,³⁶ Jordan 8.9%,⁴⁶ United Kingdom 10.3%,⁴⁷ and Western Thailand 5.9%.⁴⁸ However, the seroprevalence result of the present study is much lower than that of the previous reports in Poland 65.7%⁴⁹ and Lebanon 13.13%.⁵⁰ The higher prevalence of CAEV reported in few countries are associated with risk factors such as herd management, breed of goats, size of herd, and age of the animal.¹⁴ Also, might probably be due to unregulated trade in animals among herds.⁵⁰

It is also noted that the prevalence of the disease varies from 1.9% to 81% between countries.⁵¹ These differences in seroprevalence of CAEV among countries might be associated with the variation in the diagnostic tests, sampling method used, susceptibility of different breeds to the disease, management practices, difference in climate, and measures taken to control the disease. Also, this geographic difference in distribution of positive cases could be explained by the introduction of carrier animals from an infected area to disease free zones and the bio-security followed by farm owners.

The study also showed that sex does not have any influence on the occurrence of CAEV infection. This might be due to equal chance of access to infectious sources and susceptibility to CAEV. This finding disagrees with the report of Waseem et al,¹² Jesse et al,³⁶ and Nyi et al⁴⁸ who reported that seroprevalence was noticeably higher in females than males. This finding disagrees with Bandeira et al,⁵² who reported higher prevalence in males than females.

There was significant difference in seroprevalence among age group ($p < 0.05$), which coincided with the reports of Waseem et al,¹² Ghanem et al,¹³ Peterhans et al,¹⁹ Nyi et al,⁴⁸ and Todd.⁵³ Higher prevalence with age is best explained by horizontal transmission due to contact with CAEV infected goats. This may be because of the fact that CAEV infection is prone to infect any age of goats⁴⁶ and older animals with higher possibility to be exposed to risk factors are therefore more likely to be at risk, get infected, and remain infected for life, since CAEV is persistent and can produce lifelong infection in host.⁹ But the present finding disagrees with the finding of Jesse et al,³⁶ who reported no statistical difference between age group.

The breed related seroprevalence of CAEV infection in present study showed statistically significant difference between breeds ($p < 0.05$) and it was found that pure Boer goats were more likely to be infected followed by 50% Boer goat as compared to other breeds. Fortunately, CAEV antibody has been detected only in 3 out of 118 (2.5%) indigenous Central Highland goats which may indicate the risk of infection is largely associated with distribution of infected bucks from Ataye Boer nucleus site. In this regard, the finding of this study agreed with previously reported Tabet et al⁵⁰ and Todd.⁵³ This could be due to the indigenous breeds are more resistant and tolerant than other breeds to CAEV infection.⁸ Also, this breed susceptibility difference could be related to the influence of traits of particular family lines, the strain of the virus, and the result of one or more recessive genes.⁵⁴ There was significant variation ($p < 0.01$) between the prevalence of CAE in goats kept under different production systems. Goat kept under a semi-intensive management system was more likely to be infected than those kept under an extensive management system. This seroprevalence difference between management systems might be associated with the flock size of the farms and the housing of the animals for longer hours, mixing different breeds and age groups, and keeping a high proportion of older animals in nucleus site. Also, unfavorable housing conditions such as insufficient room, bad climatic conditions, and crowding behavior in goat might promote incidence of the disease.

Generally, the current finding indicates the existence of the diseases in goat flocks in Ethiopia. To our knowledge, this is the first report in the country, although the study conducted in limited localities and the disease might be introduced to the country with importation of live animal. Due to financial shortage, the study is conducted in some localities by using only clinical and serological test. But, it requires detailed national wide epidemiological study by using PCR/more sensitive diagnostic test to determine the overall prevalence of the diseases in small ruminant population and in order to reduce its impact on the economy of the country.

Conclusions and Recommendations

In the current study, clinical cases attributed to infection by CAEV were a major health problem; they cause production loss through morbidity and mortality in Ataye Boer nucleus site. The overall seroprevalence of CAE was 4.7%. There was significant difference ($p < 0.05$) in sero-positivity among breeds and age groups of goat; where pure Boer and 50% Boer were more affected and adults were affected at higher rate. Also, animals managed under semi-intensive were more affected than extensively managed. Apparent clinical cases prevalence attributed to CAE infection and serological result showed that the infection exists in goats flocks in Ethiopia. To the best of our knowledge, this is the first report that describes CAEV infection in Ethiopia. Further national wide epidemiological investigations need to be done to explore the seroconversion of CAEV in small ruminant and the associated management factors to enable construction of the appropriate eradication program and prevent transmission through import of live animals.

Ethical Clearance

Generally, in our research institute each proposal passes four stages of review system; Case team (animal health research case team), Directorate (Livestock research directorate), Zona (joint review by Debre birhan agricultural research centre and University), and Regional level (joint review by Amhara Regional Agricultural Livestock Research Directorate, invited International Senior Livestock researchers, and universities which have ethical committee). Finally, accepted proposals are approved by the local ethical committee called Amhara Regional Agricultural Research Institute (ARARI) Reviewer Board. So this research finding is evaluated and approved by the Amhara Regional Agricultural Research Institute Reviewer Board. This study technique followed the protocols of the National Research Ethics Review Guideline formulated by the Ministry of Science and Technology of the Federal Democratic Republic of Ethiopia in 2014. There is formal written consent between our research institute and local administrator of the study areas (Zonal, district and kebele/peasant) and also as the same time between our research institute and animal owners to do animal related research activities (breeding, nutrition and health issues) by respecting animal welfare, since the study areas are Community Based Breeding Villages. And we researchers give health service like strategic deworming against major ecto and endoparasite based on epidemiological and laboratory findings and vaccinated against major viral and bacterial disease (PPR, CCPP, sheep and goat pox, ovine

pasteurellosis, and anthrax). So, when a research team is going to do a research, only oral consent is obtained from animal owners, rather than written consents. Animals were treated with best practice of veterinary care.

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Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis, and interpretation, or in all these areas; took part in drafting, revising, or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

Disclosure

The authors report no conflicts of interest in this work.

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